

## **REMARKS**

Reconsideration of the above-identified patent application in view of the amendment above and the remarks below is respectfully requested.

No claims have been canceled or added in this paper. Claims 1, 8 and 13 have been amended in this paper. Therefore, claims 1-28 are pending and are under active consideration.

Support for the present amendment to claim 1 may be found in the present specification, for example, in the paragraph bridging pages 11 and 12, in previous claim 8, and in previous claim 13. Support for the present amendment to claim 13 may be found in claim 11, from which claim 13 depends.

Claims 1-2 and 4-28 stand rejected under 35 U.S.C. 103(a) “as being unpatentable over Eads et al. (Nucleic acids Research 2000 Vol. 28 p. 32) in view of Solinas et al. (Nucleic acids Research 2001 Vol. 29 p. e96).” In support of the rejection, the Patent Office substantially repeats its reasons of record.

Applicants respectfully traverse the subject rejection.

First, the combined teachings of the applied references fail to teach or to suggest a method that encompasses a primer falling within the scope of currently amended claim 1, i.e., a primer “whose 5’-end is joined with a probe via a linker, wherein the probe includes at least one methylation-specific CG dinucleotide, and wherein the secondary structure of the probe comprises a hairpin shape or a duplex structure” (claim 1, step b) and wherein the probe further hybridizes “intramolecularly to the primer extension product, wherein the hybridization occurs only if the cytosine positions to be analyzed were initially methylated” (claim 1, step d).

In contrast thereto, the primers described in Solinas et al. do not include a methylation-specific probe region and are not suitable for the analysis of cytosine methylation. Regarding the structure of the Scorpion primers, Solinas et al. refers to the teachings of Thelwell et al. (Nucleic Acids Research, Vol. 28(19):3752-61, 2000; see Solinas et al., page e96, left column, last paragraph). The respective Scorpion primer as described in Thelwell et al. (Thelwell et al., Table 2, page 3754) is depicted below. The underlined nucleotide stretches are those which together form the stem of the primer's stem-loop structure. The in-between nucleotides represent the probe sequence, i.e., the nucleotides which hybridize to the DNA sequence of interest:

5'-FAM CCCGCGC CTTTCCTCCACTGTTGC GCGCGGG MR HEG  
ATGGTGTGTCTTGGGATTCA-3'

(FAM - fluorescein, MR - methyl red, HEG - hexaethylene glycol, a polymerase read-through stopper)

With MethyLight, Eads et al. describes a PCR-based method for the detection of cytosine methylation. However, the combined teachings of Eads and Solinas would not have motivated a person of ordinary skill in the art to modify the Scorpion primers in such a way to analyze the methylation of cytosine positions. Instead, a person of ordinary skill in the art would have anticipated a significant risk of internal, undesired base pairings between the stem part of the Scorpion primer and a probe within the loop part that is modified to detect cytosine methylation. By said undesired base pairing, the re-assembling of the stem sequence after denaturation is prevented, whereby the fluorescent dye and the quencher molecule are spatially separated. This results in a permanent fluorescence signal, occurring independently from a proper hybridization of the probe to

its target sequence. (Applicants are currently planning to submit, within the next few weeks, a Declaration in support of the above argument.)

Accordingly, at the time the instant application was filed, a person of ordinary skill in the art would have anticipated unpredictably-occurring, false-positive signals. This is because the stem sequence of a Scorpion primer predominantly consists of GC bases (see Scorpion sequence above) for thermodynamic stability that suffices to keep the fluorescent dye and its quencher in spatial proximity. Importantly, the probe sequence of the inventive Scorpion primer, as designed for methylation analysis, as well includes “at least one methylation-specific CG dinucleotide” (claim 1, step b). Therefore, the presence of complementary sequences and base pairings between parts of the probe sequence and the stem sequence are likely to occur.

However, the combined teachings of the applied references are entirely silent about how to solve this problem. This is not remarkable since Solinas et al. refers to SNP analysis which, of course, does not require the development of methylation-specific probe sequences including CG dinucleotides. Eads et al., on the other hand, does not face this problem since the MethyLight primer oligonucleotides do not include a GC-rich stem sequence like the Scorpion primers. In summary, both references, taken individually or in combination, not only fail to teach all elements of the claimed method but also fail to provide a person of ordinary skill in the art with a suggestion or a motivation to develop the instant method.

Further, the method according to amended claim 1 would not have been obvious since the substitution of one known element (i.e., the Scorpion primer with a probe specific for a genomic DNA sequence) for another (the inventive Scorpion primer with a methylation-specific probe) would

not have yielded predictable results to one of ordinary skill in the art at the time of the invention. At that time, no methylation-specific Scorpion primers were available in the art. Accordingly, a person of ordinary skill in the art was unable to predict the internal base-pairing behavior between a GC-rich methylation-specific probe and a GC-rich sequence located in the stem structure of the primer and available to internal hybridization during every polymerase reaction cycle.

In view of these circumstances, the results of a method including the inventive methylation-specific Scorpion primers were unpredictable for a person of ordinary skill in the art at the time the invention was made, even in view of the combined teachings of the applied references. Consequently, said person of ordinary skill in the art would not have combined the elements as instantly claimed to obtain predictable results.

Accordingly, for at least the above reasons, the subject rejection should be withdrawn.

Claim 3 stands rejected under 35 U.S.C. 103(a) “as being unpatentable over Eads et al. (Nucleic acids Research 2000 Vol. 28 p. e32) in view of Solinas et al. (Nucleic acids Research 2001 Vol. 29 p. e96) as applied to Claims 1-2 and 4-28 and in further view of Berlin et al. (US Patent Application Publication 2006/0183128 August 17, 2006).” In support of the rejection, the Patent Office substantially repeats its reasons of record.

Applicants respectfully traverse the subject rejection.

Claim 3 depends from claim 1. Claim 1 is patentable over Eads et al. in view of Solinas et al. for at least the reasons given above. Berlin et al. fails to cure all of the deficiencies of Eads et al. and Solinas et al. with respect to claim 1. Therefore, based at least on its dependency from claim 1, claim 3 is patentable over the applied combination of Eads et al., Solinas et al. and Berlin et al.

Accordingly, for at least the above reasons, the subject rejection should be withdrawn.

Claims 1-28 stand provisionally rejected “on the ground of nonstatutory obviousness-type double patenting as being unpatentable over claim 1-4, 15-16, 18 of copending Application No. 11716207 in view of Solinas et al. (Nucleic acids Research 2001 Vol. 29 p. e96).”

Applicants respectfully traverse the subject rejection.

Contrary to the Patent Office’s position, U.S. Patent Application No. 11/716,207 (hereinafter “the ‘207 application”) does not render obvious the methylation-specific Scorpion primer of claim 1 of the subject application. The ‘207 application is neither drawn to the same method steps nor does it claim or suggest a primer “whose 5’-end is joined with a probe via a linker, wherein the probe includes at least one methylation-specific CG dinucleotide, and wherein the secondary structure of the probe comprises a hairpin shape or a duplex structure” (claim 1, step b) and wherein the probe further hybridizes “intramolecularly to the primer extension product, wherein the hybridization occurs only if the cytosine positions to be analyzed were initially methylated” (claim 1, step d).

The ‘207 application further fails to teach or to suggest how a Scorpion primer could be modified in order to allow the detection of cytosine methylation.

Similarly to the previous arguments made against Solinas et al. and Eads et al. that there was no motivation for a person of ordinary skill in the art to use the claimed Scorpion primer to detect cytosine methylation in DNA, the claimed subject matter of the ‘207 application, in particular, claims 1-4, 15-16, and 18, differs from the claims of the instant application, and the claims of the present invention are not coextensive in scope with the subject claims of the ‘207 application.


Accordingly, for at least the above reasons, the subject rejection should be withdrawn.

In conclusion, it is respectfully submitted that the present application is now in condition for allowance. Prompt and favorable action is earnestly solicited.

If there are any fees due in connection with the filing of this paper that are not accounted for, the Examiner is authorized to charge the fees to our Deposit Account No. 11-1755. If a fee is required for an extension of time under 37 C.F.R. 1.136 that is not accounted for already, such an extension of time is requested and the fee should also be charged to our Deposit Account.

Respectfully submitted,

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I hereby certify that this correspondence is being deposited with the United States Postal Service as first class mail in an envelope addressed to: Mail Stop RCE, Commissioner for Patents, P.O. Box 1450, Alexandria, VA 22313-1450 on 9-13-10.

  
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